

Supplementary Information

Reduction in chromosome mobility accompanies nuclear organization during early embryogenesis in *Caenorhabditis elegans*

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Table S1. *Caenorhabditis elegans* strains used in this study

Table S2. Genome sequencing of *Caenorhabditis elegans* strain CAL0872

Table S3. *lacO* spot measurement summary

Figure S1. The genomic structure of the *lacO*-inserted region in CAL0872

PacBio sequencing revealed that the *lacO* repeat was inserted on chromosome *III*. The lower diagram plots the similarity between the sequence of chromosome *III* in CAL0872 (around 3.1 Mb) and those of the WBCel235 reference genome, pKA11 plasmid (containing *lacI*::GFP), and pMK19A plasmid (containing the *lacO* repeat). The red rectangular regions indicate the positions of the *lacO* repeat, reflecting the identical sequences of the *lacO* repeat in both the CAL0872 genome and pMK19A plasmid. In CAL0872, a 5.4-kb region (3,100,175–3,105,601 bp) of *C. elegans* chromosome *III* was replaced with a 123-kb sequence containing fragments from pKA11 and pMK19A, as well as palindromic sequences derived from the *C. elegans* genome. The deleted 5.4-kb region contains the ORFs *H06I04.1* and *Y53G8B*, which are non-essential.

Figure S2. Estimation of the effect of phototoxicity: a comparison of chromosome mobility in the first and second halves of microscopic observation

To exclude the possibility that the microscopic observation procedure affected the mobility of the chromosomes, changes in the distance between the two spots in the shortest observation interval (i.e. 20 s) were compared between the first and second halves of the observation. Solid lines are from the first half and dashed lines are from the second half. The distributions of the distances were roughly Gaussian, as expected, and the variances were similar between

the two halves. Therefore, the mobility of the chromosomes was unlikely to be affected by the light illumination during microscopic observation.

Figure S3. Distribution of distances between *lacO* loci normalised by nuclear size

(a) Distributions of the distances between the two *lacO* spots for each developmental stage quantified as in Fig. 2 except that the distances were normalised by the radius of the nucleus. Different colours indicate different stages. (b) Cumulative distribution of (a).

Figure S4. Chromosomal mobility for various cell types

The MSCDs of all cells in each stage are shown (black lines). Coloured lines and dots show MSCDs of individual cell types during the 2-cell stage (orange, P1 cells; grey, AB cells), the 4-cell stage (orange, P2 cells; blue, EMS cells; grey, ABa and ABp cells), or the 8-cell stage (orange, P2 cells; purple, C cells; blue, E cells; green, MS cells; grey, ABal, ABar, ABpl, and ABpr cells).

Figure S5. Double logarithmic plot of MSCD

Double logarithmic plot version of Fig. 3a to visualise that the exponent, α , is similar among the different stages.

Movie S1. Visualization and tracking of *lacO* spots at the 2-cell stage

A representative movie for the tracking of the *lacO* spots at the 2-cell stage corresponding to Fig. 1b. Bar, 5 μm . Elapsed time is shown in min:sec.

Movie S2. Visualization and tracking of *lacO* spots at the 4-cell stage

A representative movie for the tracking of the *lacO* spots at the 4-cell stage corresponding to Fig. 1b. Bar, 5 μm . Elapsed time is shown in min:sec.

Movie S3. Visualization and tracking of *lacO* spots at the 8-cell stage

A representative movie for the tracking of the *lacO* spots at the 8-cell stage corresponding to Fig. 1b. Bar, 5 μm . Elapsed time is shown in min:sec.

Movie S4. Visualization and tracking of *lacO* spots at the 48-cell stage

A representative movie for the tracking of the *lacO* spots at the 48-cell stage corresponding to Fig. 1b. Bar, 5 μm . Elapsed time is shown in min:sec.

Table S1. *Caenorhabditis elegans* strains used in this study

Strain	Genotype	Reference
AV221	<i>unc-119(ed3) meT8 (III); meIs4 [lac-O + rol-6(su1006) + lacO] meT8 (IV); meIs1[pie-1p::GFP::lacI + unc-119(+)]</i>	Bilgir <i>et al.</i> (2013)
CAL0872	<i>unc-119 (ed3); wjIs95[pie-1p::gfp::lacI-NLS + unc-119(+)]</i> ; <i>wjIs96[lacO repeat]</i>	This study
CAL0901	<i>unc-119 (ed3); wjIs103[pie-1p::mcherry::fib-1 + unc-119(+)]</i>	This study
CAL0911	<i>unc-119 (ed3); wjIs104[pie-1p::mcherry::hpl-2 + unc-119(+)]</i>	This study
CAL0921	<i>unc-119 (ed3); wjIs105[pie-1p::15F11scFV::gfp + unc-119(+)]</i>	This study
CAL0231	<i>unc-119 (ed3); ruIs32[pie-1p::gfp::histone H2B(his-11) + unc-119(+)]III</i>	This study

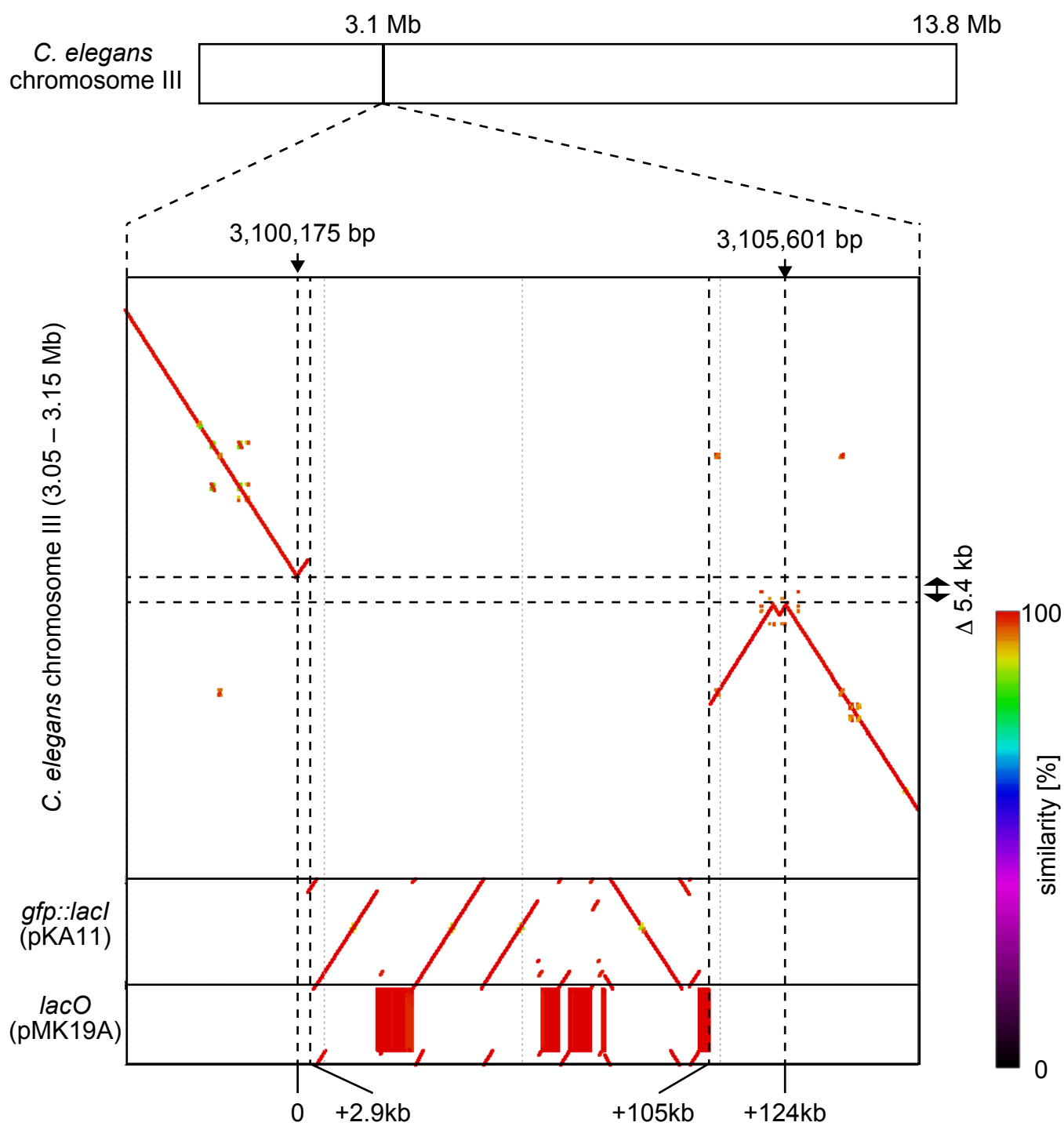
Table S2. Genome sequencing of *Caenorhabditis elegans* strain CAL0872

Data set / Assembly		Total bases (bp)	Total number	Minimum length (bp)	Maximum length (bp)	N50 (bp)	Mean (bp)
PacBio subreads		9,816,451,169	864,265	50	74,577	17,808	11,358
FALCON (≥15 kb)	Primary	101,577,359	91	18,382	5,018,309	2,274,662	1,116,014
	Alternative	439,339	15	19,293	50,678	27,673	29,289

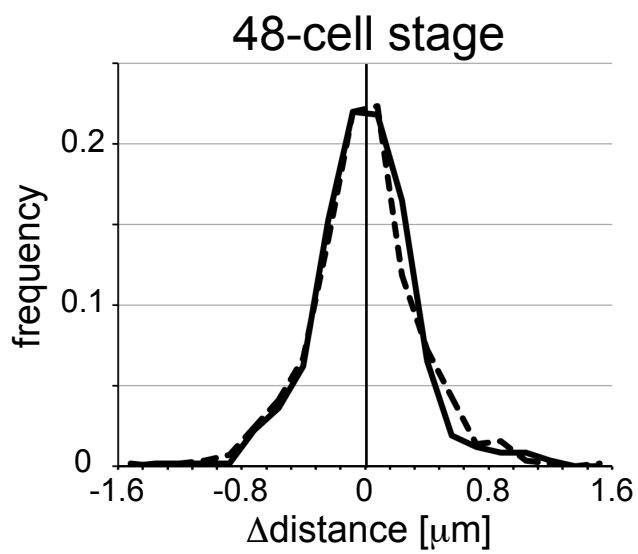
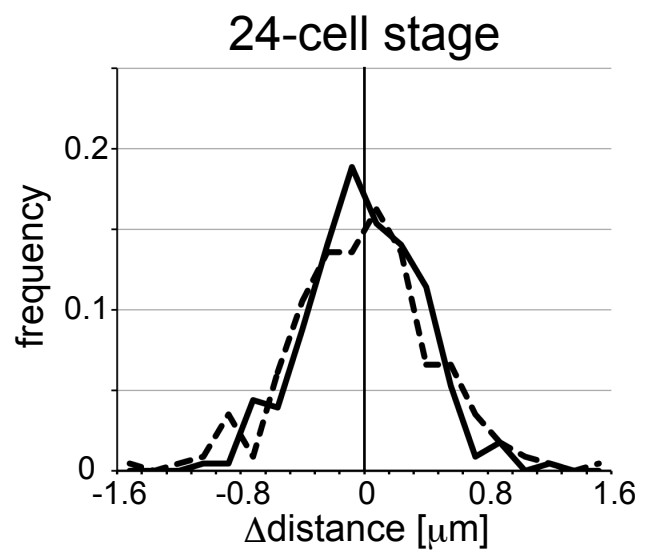
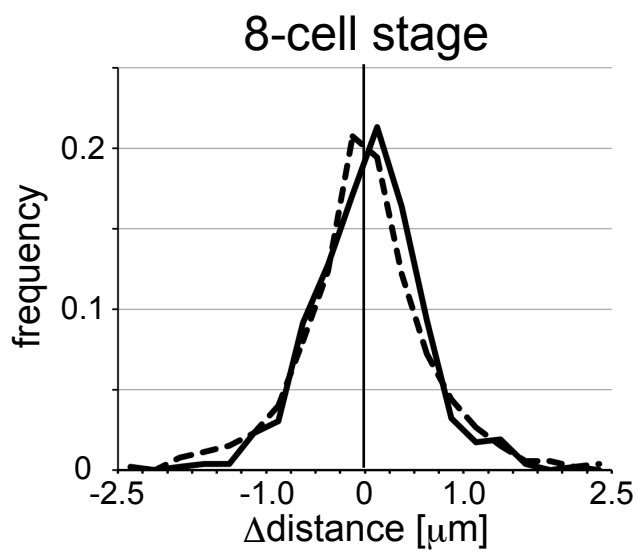
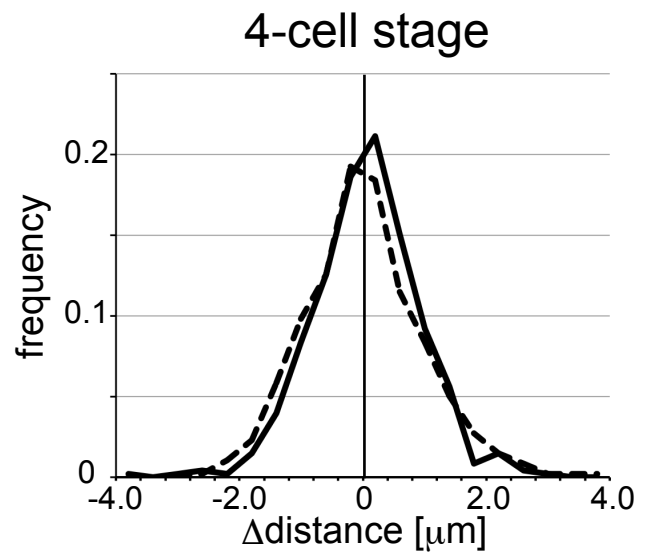
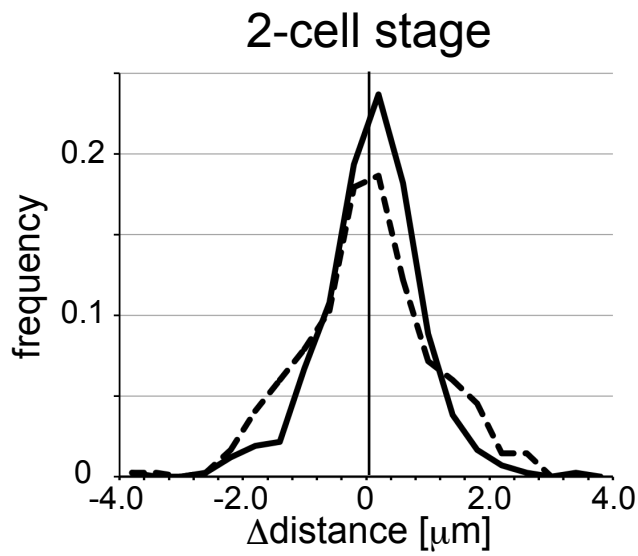
Table S3. The *lacO* spot measurement summary

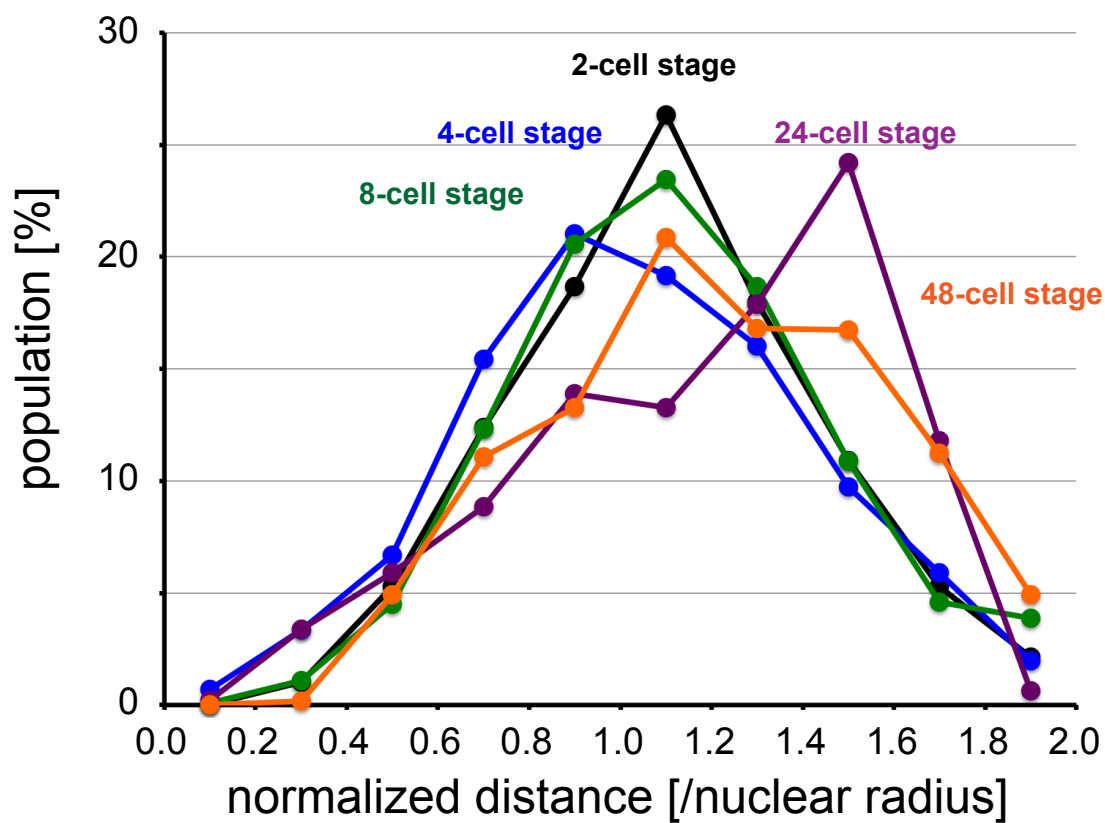
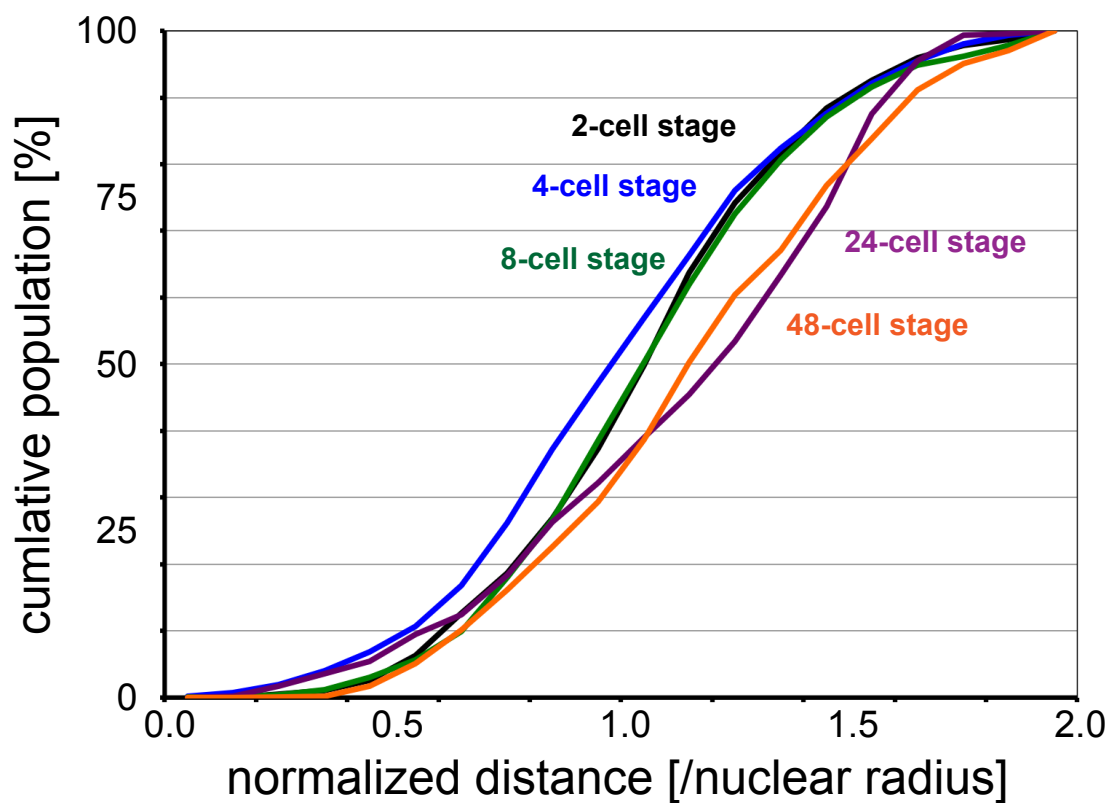
	2cell	4cell	8cell	24cell	48cell
radius [μm]	4.08	3.60	3.16	2.52	2.12
the number of nuclei	37	44	42	19	48
the number of pairs of the <i>lacO</i> spots	889	1,017	1,113	475	1,237
the total number of pairs of distance changes	10,800	11,753	14,656	5,700	15,429
the number of pairs of distance changes					
(interval = 20 s)	852	973	1,071	456	1,189
(interval =40 s)	815	929	1,029	437	1,141
(interval =60 s)	778	885	987	418	1,093
(interval =80 s)	741	841	945	399	1,045
(interval =100 s)	704	797	903	380	997
(interval =120 s)	667	753	861	361	949
(interval =140 s)	630	709	819	342	901
(interval =160 s)	593	665	777	323	853
(interval =180 s)	556	621	735	304	805
(interval =200 s)	519	577	693	285	757
(interval =220 s)	482	533	651	266	709
(interval =240 s)	445	489	609	247	661
(interval =260 s)	408	445	567	228	613
(interval =280 s)	371	401	525	209	565
(interval =300 s)	335	357	483	190	517
(interval =320 s)	302	316	441	171	469
(interval =340 s)	269	275	399	152	421
(interval =360 s)	236	239	357	133	373
(interval =380 s)	203	206	315	114	325
(interval =400 s)	172	174	273	95	277
(interval =420 s)	148	143	232	76	230
(interval =440 s)	125	115	193	57	183
(interval =460 s)	103	91	160	38	137
(interval =480 s)	82	71	132	19	93
(interval =500 s)	63	53	107		55
(interval =520 s)	52	40	86		33
(interval =540 s)	42	27	72		21
(interval =560 s)	34	16	60		12
(interval =580 s)	26	9	49		5

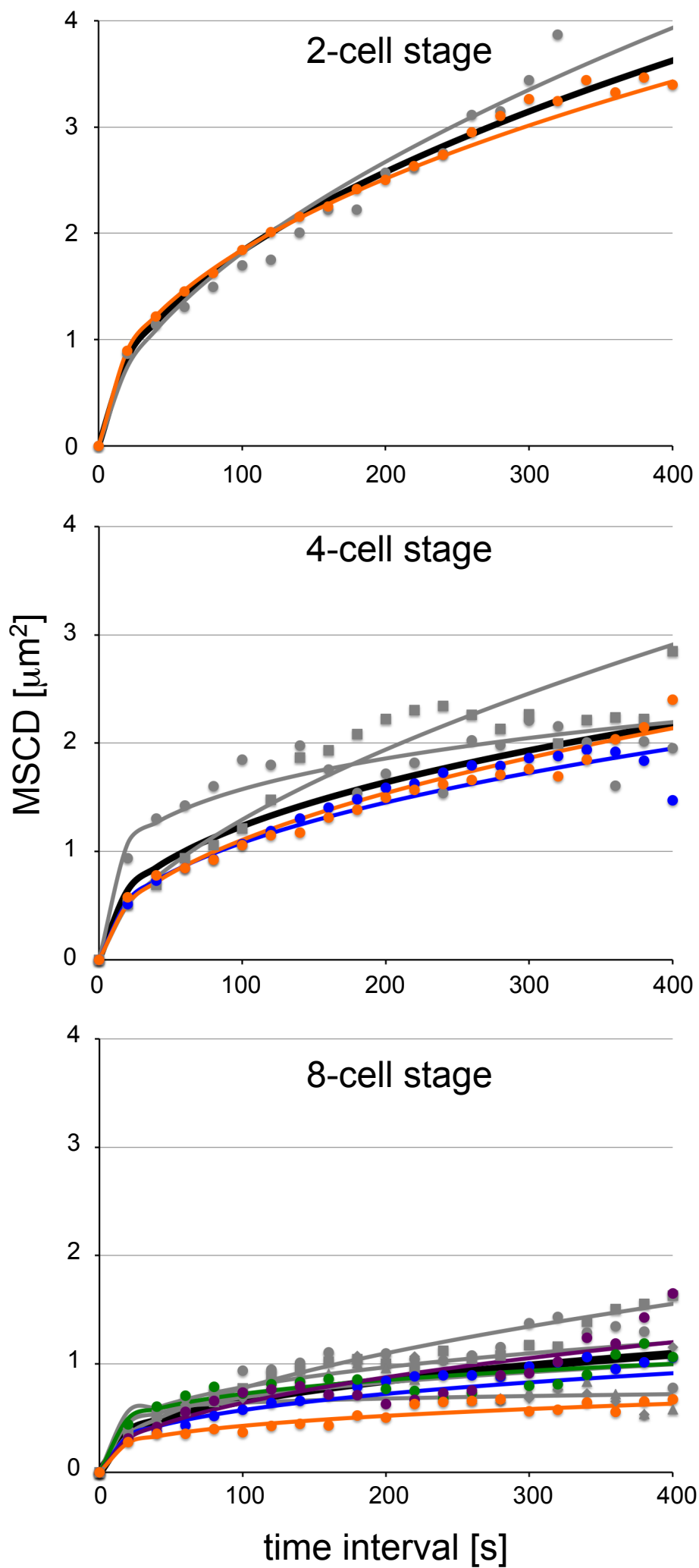
(interval =600 s)	18	3	39
(interval =620 s)	12		30
(interval =640 s)	8		23
(interval =660 s)	5		16
(interval =680 s)	3		10
(interval =700 s)	1		4
(interval =720 s)			2
(interval =740 s)			1

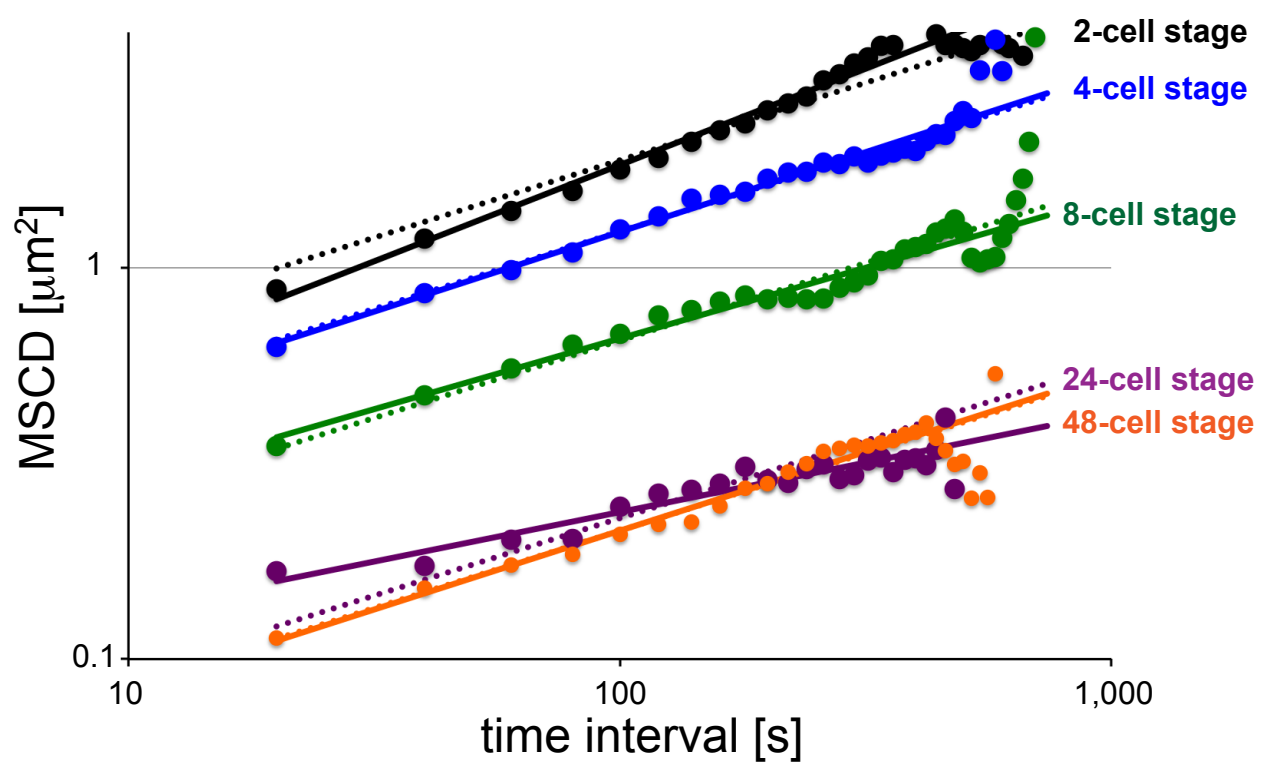


Arai et al. Figure S1



a**b**





Arai et al. Figure S5